

**THE STRUCTURE OF WAGNER'S ORGAN IN
OROPSYLLA (DIAMANUS) MONTANA MONTANA
(SIPHONAPTERA: CERATOPHYLLIDAE):
PRELIMINARY INVESTIGATIONS^{1,2}**

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ABSTRACT: The structure of Wagner's Organ in *Oropsylla (Diamanus) montana montana* was examined via light and scanning electron microscopy.

First explicitly noted by Wagner (1932a,b; 1933), many male fleas in the family Ceratophyllidae possess paired, sac-like structures lined with minute, inward directed spines, lying laterad of the base of the eighth sternum. Wagner referred to these structures as "X-glands". They are present in all but 11 of the 43 currently recognized genera in the family (Smit, 1983). Wagner and others have suggested that they have a secretory function (Wagner, 1932a,b, 1933; Günther, 1961; Traub & Rothschild, 1983), perhaps in the production of pheromones. Male Ceratophyllidae also possess well developed spiculate intersegmental membranes between the eighth and ninth segments. Wagner (1932b) has proposed that these act to provide expanded surface area to facilitate evaporation of the proposed products of the "gland". A study was begun to ascertain something more about the structure and function of these bodies, with the intention of investigating any associated secretory tissues.

MATERIALS AND METHODS

Adult male specimens of *Oropsylla (Diamanus) montana montana* (Baker, 1895), a parasite of *Spermophilus* species, were obtained from a colony maintained by the Center for Disease Control in Fort Collins, Colorado. Individuals were dissected in 2.5% glutaraldehyde and 2% paraformaldehyde in 0.1M cacodylate buffer, pH 7.3, and the tissues left in the fixative for 1-2 hours. After a buffer wash and postfixation in osmium tetroxide for 1 hour, the tissues were dehydrated in a graded ethanol series and embedded in Medcast (TM) resin. Thick sections were cut with glass

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knives, stained with toluidine blue (1% in 1% borax, aqueous), and viewed with a Zeiss photomicroscope. For observation with the scanning electron microscope, the embedded tissues were etched following the procedure of Erlandsen *et al.* (1973), sputter-coated with gold-palladium and viewed with a JEOL JSM-35 scanning microscope operated at 25KV.

RESULTS AND DISCUSSION

The arrow in Figure 1 shows the location of Wagner's Organ as it is seen in cleared specimens as they are normally mounted for study, although the location and shape vary both within and among species. In Figure 2 the organ itself with its inner spines is visible as seen in whole mounts. Figure 3 depicts a transverse section through the male abdomen, showing the relative position of the rectum (r), the aedeagus (ae), and the paired Wagner's Organs (arrows). The lumen of the organ itself is lined throughout with a thin cuticular intima, which is produced into spines of varying lengths (Fig. 4). These spines are hollow, as would be expected of this kind of cuticular growth, and many are of a length which seems out of proportion to the space within which they lie. In the dorsal region of the lumen inclusions which are spherical in section can be seen, which may be traces of a secretion associated with this organ (Fig. 5: arrow).

Most surprising is the seeming lack of glandular tissue associated with these bodies. Wagner (1932b; Fig. 2) has illustrated and discussed small cells located near the internal surface of the organ in *Ceratophyllus hirundinis*, the outer surface of which stained densely with carmine in dissected specimens. He tentatively identified these as gland cells, and the stained substance as their product. He found no connection between these cells and the lumen of the "X-gland" and suggested that the cells might lie within its walls.

During this study sections were taken well into the body of the animals, and in none of the half-dozen which were sectioned was there any clear evidence of cells or tissue of a glandular nature surrounding, or even near, the bodies. No ducts leading into the lumen of the organ were noticed in the surrounding area, the presence of which might suggest connection to a gland elsewhere in the abdomen. There is, however, as has been noted by Wagner and others in various species (Wagner, 1932b; Dampf, 1942; Traub, 1950), an opening leading from the organ to the surface of the intersegmental membrane between sterna eight and nine. Wagner's Organ itself no doubt arises as an ectodermal invagination of these membranes and opens out onto them through the "duct" thus formed.

The specimens which were sectioned were all adults when they were selected for shipment. It is possible that the production of secretions by cells

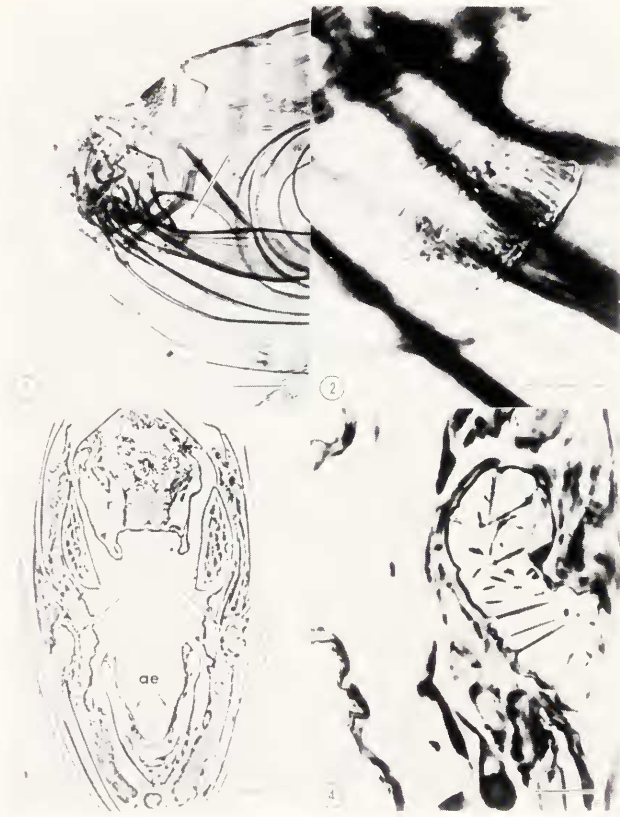


Fig. 1. Male terminalia of *Oropsylla (Diamanus) montana montana*. Location of Wagner's Organ is indicated by the arrow. Scale line = 100 microns.

Fig. 2. Light micrograph of Wagner's Organ (oil immersion). Scale line = 25 microns.

Fig. 3. Transverse section of the male abdomen, ae: aedeagus, r: rectum, arrows: Wagner's Organs. Scale line = 50 microns.

Fig. 4. Light micrograph of Wagner's Organ and surrounding tissue. Note the length of the spines. Scale line = 10 microns.

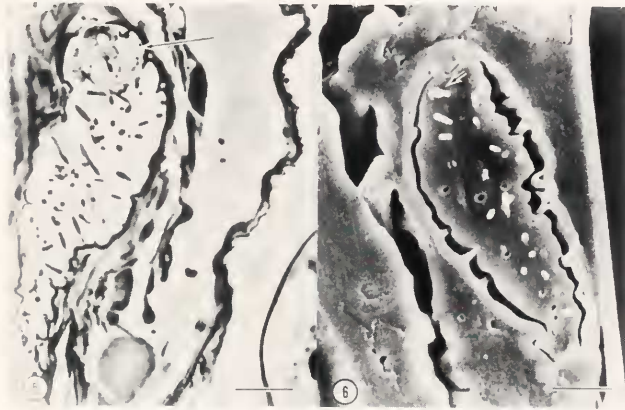


Fig. 5. As Fig. 4. Contents of the lumen are indicated by the arrow. Scale line = 10 microns.

Fig. 6. Scanning electron micrograph of resin embedded tissues. Some of the resin has been etched away to reveal the structures. The content of the lumen of the organ is undissolved resin rather than natural content. The hollow core of the spines is just visible in this micrograph (arrow). Scale line = 10 microns.

associated with this organ is dependent upon the age of the animal, and that any secretory cells present had degenerated by the time the specimens were fixed. Careful selection of males in mating condition is required to determine whether this is the case. In any event, the mode of secretion remains a puzzle, given the seeming lack of ducts leading into the organ. Subcellular cuticular ducts do occur in certain insects, for instance in the gland cells surrounding the spermathecae of some Asilidae (Reichardt, 1929; Cheetham, unpublished observations), and some such arrangement may exist here. Further work is necessary on reproductively active specimens and should include transmission electron microscopy and histochemistry.

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